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(71) Applicant: RHODIA INC. [US/US]; 259 Prospect Plains Road, Cranbury, NJ 08512 (US).

(72) Inventors: KING, William; 140 Ygnacio Court, Walnut Creek, CA 94598 (US). MING, Xintian; 307 West Oak Street, Cottage Grove, WI 53527 (US).

(74) Agents: SHEDDEN, John, A. et al.; Rhodia Inc., 259 Prospect Plains Road, Cranbury, NJ 08512 (US).

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(54) Title: HOPS ACID ANTIBACTERIAL COMPOSITIONS

WO (57) Abstract: An antibacterial composition of matter comprising: a) one or more hops acid or hops acid derivatives or hops resin or hops resin derivatives; and b) one or more food grade surfactants, surface active agents, chelating agents, antioxidants, and/or organic acids is provided.

HOPS ACID ANTIBACTERIAL COMPOSITIONS

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Field of the Invention

The present invention relates to a process for reducing the level or retarding the outgrowth of bacteria on food and nonfood products by treatment 10 with a composition which includes one or more safe and suitable hops acids or hops acid derivatives or hops resins or hops resin derivatives and one or more surfactants, surface active agents, chelators, antioxidants and/or organic acids. More specifically, the process comprises using as an ingredient or applying to a 15 food surface or a nonfood product a composition comprising beta hops acids and one or more nonionic surfactants, chelators, antioxidants and/or organic acids in order to reduce or eliminate gram positive spoilage or pathogenic bacteria, and especially strains of the harmful pathogen *Listeria monocytogenes*.

Background of the Invention

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Extensive research has been conducted in the field of food and consumer care products safety to develop compositions which function as food grade anti-bacterials. In particular, gram positive bacteria such as *Listeria monocytogenes*, *Staphylococcus aureus*, *Bacillus cereus*, *Clostridium botulinum* 25 and the like, when found in foods or consumer care products may pose significant health risks to users. Further, miscellaneous gram positive spoilage bacteria such as *lactobacilli*, *bacilli*, *streptomyces*, and *micrococci* species may cause reduction in the shelf life and appeal of processed foods and high moisture consumer care products. Both pathogenic and spoilage bacteria occur 30 most commonly at the surfaces of foods or consumer care ingredients, which

come into contact with undesirable bacteria by contamination from environmental sources. These bacteria then spread to other parts of the food through mixing, comminuting, wetting action, or migration. The bacteria may then grow during handling and storage of the product, resulting in either spoilage or infectious 5 health risks. Use of such products in sensitive body openings, such as the mouth, the skin, the eyes, or ingestion of food products profoundly increases the chance of contracting infection. In addition, other gram positive bacteria including the miscellaneous spoilage species, can opportunistically infect open cuts and wounds that are treated with consumer products normally meant for 10 external use only. The growth of all bacteria during handling and distribution must therefore be meticulously prevented and minimized in all foods and consumer care products. This invention discloses one such system for the reduction and inactivation of bacteria in food and consumer care products that are at risk.

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U.S. Patent Nos. 5,286,506 and 5,455,038 suggest that acid or acid derivatives from hops (used in beer manufacture) may have antibacterial activity against *Listeria* species. However, Johnson et al disclose in the International Journal of Food Microbiology 33 (1996) 195-207 that hops acid preparations and 20 hops acid derivatives have limited practical efficacy against bacteria in fat containing products such as cheeses, meats, sauces, and dressings. This is presumably due to the migration or entrapment of the beta acids in the fat emulsion and their subsequent unavailability for inhibition of bacterial growth in the aqueous portion of the emulsion where bacteria are known to grow. This 25 problem of poor activity of hops beta acids in fatty acid containing foods has prevented them from being used commercially as natural anti-microbial agents for control of listeria or other gram positive pathogens. Further, it is unlikely that hops acid activity would prove effective in the water in oil emulsions common among most consumer care products. The ability to tag the hops acids in the 30 aqueous phase of such systems is a property that would dramatically increase

their value as anti-microbials in either food or consumer care products containing high oil or fatty acid levels.

To the extent necessary for completion of this patent application, all of 5 the cited references are expressly incorporated by reference. However, despite the above teachings, there still exists a need in the art for a method for protecting fat containing foods and consumer care products against bacterial growth using natural, generally recognized as safe (GRAS) substances. More specifically, there exists a need for more complete and effective reduction of harmful 10 pathogens by use of safe and suitable levels of food grade anti-microbial ingredients such as hops acids whereby the hops acids may be distributed effectively to the aqueous phase of the products to be treated.

Brief Summary of the Invention

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It is now discovered, quite surprisingly, that a composition which has as a first component, at least one compound selected from the group consisting of hops acids, hops acid derivatives, hops resins, and hops resin derivatives; and as a second component, at least one compound selected from the group 20 consisting of food grade surfactants, surface active agents, chelators, antioxidants, and organic acids provides excellent antibacterial properties, especially against potentially harmful gram positive bacteria of the *Listeria* genus and can easily be applied to the foods or consumer products based on oil in water emulsions.

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One embodiment of the present invention comprises an antibacterial composition comprising: (a) a first component having at least one compound selected from the group consisting of hops acids, hops acid derivatives, hops resins, and hops resin derivatives; and (b) a second component having at least 30 one compound selected from the group consisting of food grade surfactants and surface active agents. Particularly preferred is a composition containing beta

hops acids and a food grade surfactant or surface agent which is either a nonionic surfactant, a propylene glycol, or a mixture thereof.

In another embodiment, the present invention provides a method for reducing, 5 retarding, or even eliminating gram positive bacteria, and especially the *Listeria monocytogenes*, in food products comprising the step of treating the surfaces of said food product with a bacteriostatically or bactericidally effective amount of a composition comprising: (a) a first component having at least one compound selected from the group consisting of hops acids, hops acid derivatives, hops 10 resins, and hops resin derivatives; and (b) one or more food grade surfactants and/or surface active agents.

It is an object of the present invention to provide a process for treating food products in order to protect against harmful bacteria.

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An additional object of the present invention is to provide a novel composition having substantially greater antibacterial properties than previously observed for the individual components of the composition.

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An additional object of the present invention is to provide a product which is easily applied to all surfaces of a food product which is potentially susceptible to bacterial degradation.

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A final object of the present invention is to provide a product which can work well under the partitioning conditions of a fat or oil in water emulsion.

These, and other objects, will readily be apparent to those skilled in the art as reference is made to the detailed description of the preferred embodiment.

30 Detailed Description of the Preferred Embodiment

In describing the preferred embodiment, certain terminology will be utilized for the sake of clarity. Such terminology is intended to encompass the recited embodiment, as well as all technical equivalents which operate in a similar manner for a similar purpose to achieve a similar result.

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The present invention provides a novel antibacterial composition and its use in a process for reducing, retarding, or totally eliminating harmful bacteria, preferably from food surfaces.

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The novel antibacterial composition comprises: (a) one or more hops acids or hops acid derivatives or hops resins or hops resin derivatives or mixtures thereof; and (b) one or more food grade surfactants or surface active agents or chelators, or antioxidants, or organic acids, or mixtures thereof, preferably food grade surfactants or surface active agents or mixtures thereof.

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The first component of the novel composition is one or more hops acids, or hops acid derivatives, or hops resins, or hops resin derivatives, or mixtures thereof. The bitter acids component of the hops used in beer making and particularly the beta-acids have now been found to be useful as bactericides in food products. The most prevalent groups of bitter acids found as components of hops are the alpha-acids and the beta-acids, also referred to as humulones and lupulones, respectively. Both contribute bitterness to beer, but the alpha-acids are much more intense in this regard than the beta-acids. Producers of hop extracts isolate the alpha and beta acids commercially by various chromatographic techniques and have recently developed a technique to separate the two acid fractions using liquid carbon dioxide under supercritical conditions. A by-product of the operation is a product which contains approximately 61 weight percent beta-acids, the remainder consisting essentially of hops resins. This by-product can be standardized with malto-dextrin or other food grade carrier, spray-dried, and used as an antibacterial food ingredient. A

preferred beta hops acids component is commercially available as a natural flavor extract containing 1 weight percent beta hops acids.

The alpha-acids contained in hops are commonly known as humulone, 5 cohumulone and adhumulone, while the beta-acids contained in hops are commonly known as lupulone, colupulone and adlupulone. Derivatives of the hops acids or hops resins which have demonstrated antibacterial properties such as hexahydrocolupulone and tetrahydroisohumulone, as disclosed in U.S. Patent No. 5,455,038, are specifically contemplated for use in association with the 10 present invention. Also considered as specifically contemplated for use in association with the present invention is the use of the acid salt forms of the hops acids or hops resins.

In practice, the hop acid or resin or derivative thereof is added to the food 15 product in amounts between about 0.1 to about 50 ppm (by weight of solution used for treatment), more preferably between about 0.40 to about 20 ppm.

The preferred second component of the novel composition comprises 20 one or more food grade surfactants or surface active agents. The term "surface active agent" is intended to also include what is commonly known as a protective colloid.

The surfactant employed preferably is a food grade emulsifier with a high 25 hydrophilic/lipophilic balance (HLB) value. Such values should range from about 10 to about 16. Suitable emulsifiers preferably have also been approved for use in foods since many of the applications for the present invention reside in the food and drug industries. Preferably, the emulsifier is selected from the group consisting of polyoxyethylene sorbitan esters, sorbitan esters, monoglycerides, 30 diglycerides, lecithin, polyglycerols, sodium stearoyl-2-lactylate, stearyl-2-lactyl acid, polyoxystearates, acetylated monoglycerides and mixtures thereof. Preferably, sorbitan esters and polyoxyethylene sorbitan esters are the

emulsifiers of choice. The above list is considered only a representative listing as the present invention contemplates the use of any food grade surfactant.

Particularly preferred commercially available surfactants include: Tween-5 80, a trademark of ICI Americas Inc. for polyoxyethylene sorbitol ester; Tween-20, a trademark of ICI Americas Inc. for polyoxyethylenesorbitan monolaurate; and Triton X-100, a trademark of Union Carbide Chemicals and Plastics Co. Inc. for octylphenol ethylene oxide condensate.

10 When a surfactant is used in combination with the hops acids or resins or hops acid or resin derivatives, it is added in an amount ranging from about 0.1 to 10%, more preferably between 0.5 to 5% of the treatment composition.

15 Alternatively, a food grade surface active agent other than a surfactant can be selected. This includes, but is not limited to polyhydric alcohols such as polyethylene glycols, more specifically polypropylene glycol, and other compounds having multiple hydroxy groups such as, glycerol, sorbitol, mannitol, inositol, and the like. Protective colloids such as polyvinyl alcohol are also specifically considered for use in accordance herewith.

20 When a surface active agent other than a surfactant is used in combination with the hops acids or hops acid derivatives, it is added in an amount ranging from about 0.1 to 20%, more preferably between 0.5 to 10% of the treatment composition.

25 The combination of hops acids or hops acid derivatives with both a surfactant and a surface active agent other than a surfactant is also specifically contemplated as falling within the scope of the present invention.

30 The second component of the compositions of this invention can comprise a chelating agent; preferably one selected from the group consisting of

citrates, disodium ethylenediaminetetraacetate (Na₂ EDTA), dicalcium ethylenediaminetetraacetate (CaEDTA), ethylene glycol-bis(beta-aminoethyl ether)-tetraacetic acid (EGTA), and mixtures thereof. Most preferably, the chelating agent comprises sodium citrate.

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Alternatively, the second component of the compositions of this invention can comprise an antioxidant; preferably one selected from the group consisting of butylated hydroxyanisole, butylated hydroxytoluene, mono-tertbutylhydroquinone (TBHQ), propylgallate ascorbic acid, tocopherols, and mixtures thereof. Most preferably, the antioxidant comprises butylated hydroxyanisole.

10 Also, the second component of the compositions of this invention can comprise an organic acid; preferably one selected from the group consisting of acetic acid, lactic acid, propionic acid, benzoic acid, sorbic acid, and their salts.

15 Other additives which can be present in the inventive composition include, but are not limited to the following materials: other antibacterial agents, natural or synthetic seasonings, oils, and/or flavors, dyes and/or colorants, 20 vitamins, minerals, nutrients, enzymes, binding agents such as guar gum and xanthan gum and the like. The addition of these materials is not considered critical to the success of the present invention and this addition would be considered to be within the skill of the artisan.

25 The antimicrobial composition of the present invention may be used in connection with any food or high moisture consumer care product that is susceptible to bacterial growth or degradation. Typical food products include, but are not limited to fruits and vegetables including derived products, grain and grain derived products, dairy foods, cooked meats, poultry, and seafood. In 30 particularly preferred embodiments, the composition is used in connection with meat, poultry and/or seafood, home meal replacements, soups, deli salads,

processed cheese, tofu, beverages, or other ready to eat food products. Typical consumer care applications include, but are not limited to moisture containing cosmetics, oral care products, skin care products, hair care products, eye care products, feminine hygiene products, liquid soaps, shampoos, and detergents.

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To selectively reduce the level of gram positive bacteria on a food surface, the novel composition may be applied to the food surface. In practice the application of the composition of matter to the food surface may either be a direct application or an indirect application such as by first coating a packaging material or casing and subsequently bringing the packaging material or casing into intimate contact with the food surface. The use of the term "food surface" is defined to include any and all internal or external surfaces of the food product being treated.

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The composition according to the present invention is most readily used by mixing along with the other ingredients in a blended food or consumer care product, but should also be effective to treat the surface of solid food products by a rinse, spray, packaging material, or by application to the interior of such products, e.g., by injection. In still other embodiments, the composition may be applied as a marinate, breading, seasoning rub, glaze, colorant mixture, and the like, the key criteria being that the antimicrobial composition be available to the surface subject to bacterial degradation. In still other embodiments, the composition may be indirectly placed into contact with the food surface by applying the composition to food packaging and thereafter applying the packaging to the food surface. The optimum amount to be used will depend on the composition of the particular food product to be treated and the method used for applying the composition to the food surface, but can be determined by simple experimentation.

30

The compositions of this invention are effective against gram positive bacteria including, but not limited to the following bacteria: *Listeria* bacteria such

as *Listeria monocytogenes*, *Staphylococcus* bacteria such as *Staphylococcus aureus*, *Clostridia* bacteria, *Bacillus* bacteria, micrococci, streptococci, or other lactic acid bacteria. Under certain conditions, use of the inventive composition reduces bacterial levels to levels undetectable by standard enrichment 5 techniques.

The invention will be further described by the following non-limiting examples.

10 Example 1

Activity of Beta Hop Acid (BHA) and a non-ionic surfactant against *Listeria monocytogenes*

15 As shown in Table 1, two groups of tests are conducted in Trypticase soy broth, pH 6.0 at 30°C for 48 hours to show the effect of a bactericidal system containing BHA and the surfactant Tween-80 (polyoxyethylene sorbitol ester). The test of BHA without Tween-80 serves as the control to show the effect of BHA alone against *L. monocytogenes*. The test of BHA with Tween-80 20 demonstrates a surprisingly enhanced bactericidal activity of this composition against *L. monocytogenes*. Table 1 shows that at a concentration of 0.125 ppm BHA and 2% Tween-80, the bactericidal system has a 3 log reduction of the target bacteria, while less than 1 log deduction is observed at the same concentration of BHA without Tween-80. At higher concentrations of BHA, the 25 BHA and Tween-80 combination virtually eliminates the bacteria levels to less than 10/ml. Thus, the composition of BHA and Tween-80 demonstrate a synergistic activity of greater than 20 times that of BHA alone.

Table 1

5	BHA preparation *	CFU/ml	
		with 2% Tween-80	without Tween-80
		ppm	
10	0	2.2 x 10e8	2.1 x 10e8
	250 (0.125)	1.5 x 10e4	3.3 x 10e7
	500 (0.25)	20	5.4 x 10e6
	1000 (0.5)	<10	4.6 x 10e6
	5000 (2.5)	<10	1.7 x 10e6

* BHA used is a raw extract preparation that contains about 5000 ppm pure beta
15 hop acids.

Example 2

Activity of BHA and a water activity reducing agent against *Listeria*
20 *monocytogenes*

As shown in Table 2, two groups of tests are conducted in Trypticase soy
broth, pH 6.0 at 30°C for 48 hours to show the effect of the bactericidal system
containing BHA and propylene glycol, a water activity reducing agent. The test of
25 BHA without propylene glycol served as the control to show the effect of BHA
alone against *L. monocytogenes*. The test of BHA with propylene glycol
demonstrates surprisingly enhanced bactericidal activity of the composition
against *L. monocytogenes*. Table 2 shows that at a concentration of 0.125 ppm
BHA and 8% propylene glycol, the bactericidal system causes a 2.5 log
30 reduction in the target bacteria, while less than 1 log reduction is observed at the
same concentration of BHA without propylene glycol. At higher concentrations of

BHA, the BHA and propylene glycol combination virtually eliminate the bacteria, i.e., the combination reduces the levels to less than 10/ml. Thus, the composition of BHA and propylene glycol demonstrates a synergistic activity enhancement of greater than 20 times that of BHA alone.

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Table 2

	BHA preparation *	CFU/ml	
		with 8% propylene glycol (PG)	without PG
10	ppm		
	0	2.2 x 10e8	2.1 x 10e8
	250 (0.125)	7.8 x 10e5	3.3 x 10e7
	500 (0.25)	8.1 x 10e4	5.4 x 10e6
	1000 (0.5)	1000	4.6 x 10e6
15	5000 (2.5)	< 10	1.7 x 10e6

* BHA used is a raw extract preparation that contains about 5000 ppm pure beta hop acids.

20

Example 3

Synergism of Beta Hop Acid (BHA) and a chelating agent against *Listeria monocytogenes*

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As shown in Table 3, two groups of tests are conducted in trypticase soy broth, pH 6.0 at 30°C for 48 hours to show the effect of a bactericidal system containing BHA preparation and a chelating agent, sodium citrate. The test of BHA without citrate serves as the control to show the effect of BHA alone against *L. monocytogenes*. The test of BHA with chelating agent demonstrates a surprising enhanced bactericidal activity of this composition against *L.*

monocytogenes. Table 1 shows that at a concentration of 50 ppm BHA preparation and 0.5% sodium citrate, the bactericidal system has a 6 log reduction against the target bacteria, while less then 4 log reduction is observed at the same concentration of BHA preparation. Thus, the composition of BHA preparation and a chelating agent demonstrate a synergistic effect against *L. monocytogenes*.

Table 3

10	BHA preparation *	CFU/ml	
		with 0.5% NaCitrate	without NaCitrate
0		2.0 x 10e8	2.1 x 10e8
50 ppm		40	6 x 10e4

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* beta hop acid preparation contains about 0.97% pure beta hop acids.

Example 4

20 Synergism of Beta Hop Acid (BHA) and antioxidant against *Listeria monocytogenes*

As shown in Table 4, two groups of tests are conducted in Trypticase soy broth, pH 6.0 at 30°C for 48 hours to show the effect of the bactericidal system containing BHA preparation and butylated hydroxyanisole, an antioxidant. The test of BHA without antioxidant served as the control to show the effect of BHA alone against *L. monocytogenes*. The test of BHA preparation with butylated hydroxyanisole demonstrates surprisingly enhanced bactericidal activity of the composition against *L. monocytogenes*. Table 2 shows that at a concentration of 30 50 ppm BHA preparation and 100 ppm butylated hydroxyanisole, the bactericidal composition causes a 6 log reduction in the target bacteria, while less then 4 log

reduction is observed at the same concentration of BHA preparation without butylated hydroxyanisole. Thus, the composition of BHA preparation and an antioxidant demonstrates a synergistic effect against *L. monocytogenes*.

5

Table 4

		CFU/ml	
BHA preparation *		with 100 ppm butylated hydroxyanisole	without butylated hydroxyanisole
10	0	1.2 x 10e8	2.1 x 10e8
	50 ppm	50	6 x 10e4

* beta hop acid preparation contains about 0.97% pure beta hop acids.

15 Example 5

Synergism of Beta Hop Acid (BHA) and organic acids against *Listeria monocytogenes*

20 As shown in Table 5, two groups of tests are conducted in Trypticase soy broth, pH 6.0 at 30°C for 48 hours to show the effect of the bactericidal composition containing BHA preparation and organic acids : acetic acid, propionic acid and lactic acids. The test of BHA preparation served as the control to the synergistic effect against *L. monocytogenes*. The test of BHA preparation with organic acids demonstrates enhanced bactericidal activity of the composition against *L. monocytogenes*. Table 3 shows that at a concentration of 50 ppm BHA preparation and 500 ppm organic acids, the bactericidal composition causes a 3 log reduction in the target bacteria, while less than 4 log reduction is observed at the same concentration of BHA preparation when used alone. Thus, the composition of BHA preparation and an organic acid demonstrates a synergistic effect against *L. monocytogenes*.

Table 5

5	BHA preparation *	<u>CFU/ml</u>		
		<u>with 500 ppm organic acid</u>		
		<u>acetic</u>	<u>propionic</u>	<u>lactic</u>
	0	1.2 x 10e8	1.4 x 10e8	2.0 x 10e8
	50 ppm	1.5 x 10e3	2.0 x 10e3	4.2 x 10e3
				6 x 10e4

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* beta hop acid preparation contains about 0.97% pure beta hop acids.

Having described the invention in detail and by reference to the preferred embodiments thereof, it will be apparent that modifications and variations are possible without departing from the scope of the appended claims.

WHAT IS CLAIMED IS:

1. An antibacterial composition of matter comprising: (a) a first component including at least one compound selected from the group consisting of hops acids, hops acid derivatives, hops resins, and hops resin derivatives; and (b) a second component including at least one compound selected from the group consisting of food grade surfactants, surface active agents, chelating agents, antioxidants and organic acids.
- 10 2. An antibacterial composition of matter comprising: (a) a first component including at least one compound selected from the group consisting of hops acids, hops acid derivatives, hops resins, and hops resin derivatives; and (b) a second component including at least one compound selected from the group consisting of food grade surfactants and surface active agents.
- 15 3. The composition according to claim 2 wherein said second component comprises a nonionic surfactant.
- 20 4. The composition according to claim 2 wherein said second component is selected from the group consisting of polyoxyethylene sorbitan esters, sorbitan esters, monoglycerides, diglycerides, lecithin, polyglycerols, sodium stearoyl-2-lactylate, stearyl-2-lactyllic acid, polyoxystearates, acetylated monoglycerides and mixtures thereof.
- 25 5. The composition according to claim 1 wherein said second component comprises a polyhydric alcohol.
- 30 6. The composition according to claim 5 wherein said polyhydric alcohol is selected from the group consisting of polyethylene glycols, glycerol, sorbitol, mannitol, inositol and mixtures thereof.

7. The composition according to claim 1 wherein said second component comprises both a surfactant and a surface active agent other than a surfactant.
- 5 8. The composition according to claim 1 wherein said second component comprises a chelating agent.
9. The composition according to claim 8 wherein said chelating agent is selected from the group consisting of citrates, disodium 10 ethylenediaminetetraacetate (Na₂ EDTA), dicalcium ethylenediaminetetraacetate (CaEDTA), ethylene glycol-bis(beta-aminoethyl ether)-tetraacetic acid (EGTA), and mixtures thereof.
10. The composition according to claim 8 wherein said chelating agent is 15 sodium citrate.
11. The composition according to claim 1 wherein said second component comprises an antioxidant.
- 20 12. The composition according to claim 11 wherein said antioxidant is selected from the group consisting of butylated hydroxyanisole, butylated hydroxytoluene, mono-tertbutylhydroquinone (TBHQ), propylgallate ascorbic acid, tocopherols, and mixtures thereof.
- 25 13. The composition according to claim 12 wherein said antioxidant comprises butylated hydroxyanisole.
14. The composition according to claim 1 wherein said second component comprises an organic acid.

15. The composition according to claim 14 wherein said organic acid is selected from the group consisting of acetic acid, lactic acid, propionic acid, benzoic acid, sorbic acid, and their salts.
- 5 16. The composition according to claim 1 wherein the amount of the first component by weight of the composition is 0.1 to 500 ppm.
17. The composition according to claim 2, wherein the amount of said surfactant is 0.1 to 10% by weight of the composition.
- 10 18. The composition according to claim 2, wherein the amount of said surfactant is 0.5 to 5% by weight of the composition.
- 15 19. The composition according to claim 2 wherein the amount of said surface active agent is 0.1 to 20% by weight of the composition.
- 20 20. The composition according to claim 2 wherein the amount of said surface active agent is 0.5 to 10% by weight of the composition.
- 20 21. The composition according to claim 1 wherein the amount of said chelating agent is 0.01 to 5% by weight of the composition.
- 25 22. The composition according to claim 1 wherein the amount of said chelating agent is 0.1 to 1% by weight of the composition.
- 25 23. The composition according to claim 1 wherein the amount of said antioxidant is 50 to 1000 ppm of the composition.
- 30 24. The composition according to claim 1 wherein the amount of said antioxidant is 100 to 500 ppm of the composition.

25. The composition according to claim 1 wherein the amount of said organic is 0.01 to 5% by weight of the composition.

26. The composition according to claim 1 wherein the amount of said 5 organic is 0.05 to 1 % of the composition.

27. A method for reducing gram positive bacteria in food and non food products comprising the step of treating the products with a bacteristatically or bactericidally effective amount of a composition comprising: (a) a first 10 component including at least one compound selected from the group consisting of hops acids, hops acid derivatives, hops resins, and hops resin derivatives; and (b) a second component including at least one compound selected from the group consisting of food grade surfactants, surface active agents, chelating agents, antioxidants and organic acids.

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28. A method for reducing gram positive bacteria in food and non food products comprising the step of treating the products with a bacteristatically or bactericidally effective amount of a composition comprising: (a) a first 20 component including at least one compound selected from the group consisting of hops acids, hops acid derivatives, hops resins, and hops resin derivatives; and (b) a second component including at least one compound selected from the group consisting of food grade surfactants and surface active agents.

29. The method of claim 28 wherein said second component comprises a 25 nonionic surfactant.

30. The method of claim 28 wherein said second component is selected from the group consisting of polyoxyethylene sorbitan esters, sorbitan esters, monoglycerides, diglycerides, lecithin, polyglycerols, sodium stearoyl-2-30 lactylate, stearyl-2-lactyllic acid, polyoxystearates, acetylated monoglycerides and mixtures thereof.

31. The method of claim 27 wherein said second component comprises a polyhydric alcohol.

32. The method of claim 31 wherein said polyhydric alcohol is selected from 5 the group consisting of polyethylene glycols, glycerol, sorbitol, mannitol, inositol and mixtures thereof.

33. The method of claim 28 wherein said second component comprises both a surfactant and a surface active agent other than a surfactant.

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34. The method of claim 27 wherein said second component comprises a chelating agent.

35. The method of claim 34 wherein said chelating agent is selected from 15 the group consisting of citrates, disodium ethylenediaminetetraacetate (Na₂ EDTA), dicalcium ethylenediaminetetraacetate (CaEDTA), ethylene glycol-bis(beta-aminoethyl ether)-tetraacetic acid (EGTA), and mixtures thereof.

36. The method of claim 35 wherein said chelating agent is sodium citrate.

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37. The method of claim 27 wherein said second component comprises an antioxidant.

38. The method of claim 37 wherein said antioxidant is selected from the 25 group consisting of butylated hydroxyanisole, butylated hydroxytoluene, mono-tertbutylhydroquinone (TBHQ), propylgallate ascorbic acid, tocopherols, and mixtures thereof.

39. The composition according to claim 38 wherein said antioxidant 30 comprises butylated hydroxyanisole.

40. The method of claim 27 wherein said second component comprises an organic acid.
- 5 41. The method of claim 40 wherein said organic acid is selected from the group consisting of acetic acid, lactic acid, propionic acid, benzoic acid, sorbic acid, and their salts.
- 10 42. The method of claim 27 wherein said food product is selected from the group consisting of meat, poultry, seafood, home meal replacements, soups, deli salads, processed cheese, tofu, beverages, other ready to eat food products, and mixtures thereof.
- 15 43. The method according to claim 27 wherein said product comprises an oil in water emulsion.
- 20 44. The method of claim 27 wherein said nonfood product is selected from the group consisting of moisture containing cosmetics, oral care products, skin care products, hair care products, eye care products, feminine hygiene products, liquid soaps, shampoos, and detergents.
- 25 45. The method of claim 27 wherein the bacterial levels of any one or more of the following bacteria are reduced to levels undetectable by standard enrichment techniques: *Listeria* bacteria, *Staphylococcus* bacteria, *Clostridia* bacteria, *Bacillus* bacteria, or lactic acid bacteria.

INTERNATIONAL SEARCH REPORT

Internat' Application No
PCT/US 00/20484

A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 A23L3/3472 A23L3/3508

According to International Patent Classification (IPC) or to both national classification and IPC.

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC 7 A23L A23B

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

PAJ, EPO-Internal, WPI Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	US 4 170 638 A (OWADES JOSEPH L) 9 October 1979 (1979-10-09)	1-3, 5, 6, 14, 17, 18, 27, 28, 31, 32, 40, 44, 45 38 42
Y	column 3, line 16 - line 25; claims 1, 11; example 2 column 1, line 26 -column 60 ----	
Y	WO 99 08547 A (COURT NOREEN BERNADETTE ;COURT ROBERT EDWARD (IE); MALTEX LIMITED) 25 February 1999 (1999-02-25) claims 1, 3, 4, 12 ----	38 -/-

Further documents are listed in the continuation of box C.

Patent family members are listed in annex.

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Date of the actual completion of the international search

Date of mailing of the international search report

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Name and mailing address of the ISA
European Patent Office, P.B. 5818 Patentlaan 2
NL - 2280 HV Rijswijk
Tel. (+31-70) 340-2040, Tx. 31 651 epo nl.
Fax: (+31-70) 340-3016

Authorized officer

Guyon, R

INTERNATIONAL SEARCH REPORT

Internat: Application No

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C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	US 5 455 038 A (BARNEY MICHAEL C ET AL) 3 October 1995 (1995-10-03) column 1, line 20 -column 2, line 31; claims ---	42
X	PATENT ABSTRACTS OF JAPAN vol. 018, no. 367 (C-1223), 11 July 1994 (1994-07-11) & JP 06 098738 A (ASAMA KASEI KK), 12 April 1994 (1994-04-12) abstract ---	1-7, 11, 14, 15, 27-29, 31, 33, 40-42
X	US 5 166 449 A (TODD JR PAUL H ET AL) 24 November 1992 (1992-11-24)	1-3, 5, 6
Y	claims; examples ---	27, 28
Y	US 5 286 506 A (MILLIS JAMES R ET AL) 15 February 1994 (1994-02-15) claims 1, 4, 5 ---	27, 28
Y	PATENT ABSTRACTS OF JAPAN vol. 1995, no. 11, 26 December 1995 (1995-12-26) & JP 07 196572 A (ASAHI BREWERIES LTD), 1 August 1995 (1995-08-01) abstract ---	1
Y	US 5 578 307 A (WUNDERLICH JENS-CHRISTIAN ET AL) 26 November 1996 (1996-11-26) column 3, line 63 -column 4, line 20; claims; examples ---	1
A	WO 97 33971 A (KALAMAZOO HOLDINGS INC) 18 September 1997 (1997-09-18) page 2, line 23 - line 32; claims 1, 8, 24, 25 -----	1, 34, 35

INTERNATIONAL SEARCH REPORT

Information on patent family members

Internat' l Application No

PCT/US 00/20484

Patent document cited in search report		Publication date		Patent family member(s)		Publication date
US 4170638	A	09-10-1979	DE	2749274 A		11-05-1978
WO 9908547	A	25-02-1999	AU	8745998 A		08-03-1999
			EP	1003385 A		31-05-2000
US 5455038	A	03-10-1995	US	5370863 A		06-12-1994
			CA	2111375 A		17-06-1994
			EP	0606599 A		20-07-1994
JP 06098738	A	12-04-1994		NONE		
US 5166449	A	24-11-1992	US	4918240 A		17-04-1990
			US	5082975 A		21-01-1992
			AU	619143 B		16-01-1992
			AU	3958889 A		15-02-1990
			BE	1004239 A		20-10-1992
			CA	1315804 A		06-04-1993
			DE	3926332 A		22-02-1990
			GB	2222160 A, B		28-02-1990
			NL	8902050 A, B		01-03-1990
			ZA	8906186 A		24-04-1991
US 5286506	A	15-02-1994	AU	5411194 A		24-05-1994
			CA	2147646 A, C		11-05-1994
			EP	0668756 A		30-08-1995
			JP	2731632 B		25-03-1998
			JP	8502887 T		02-04-1996
			KR	153063 B		01-10-1998
			NZ	257479 A		21-12-1995
			WO	9409759 A		11-05-1994
JP 07196572	A	01-08-1995		NONE		
US 5578307	A	26-11-1996	DE	4201172 C		22-07-1993
			DE	4201179 A		22-07-1993
			AT	151631 T		15-05-1997
			AU	679905 B		17-07-1997
			AU	3343193 A		03-08-1993
			CA	2128242 A, C		22-07-1993
			WO	9313754 A		22-07-1993
			DE	59306197 D		22-05-1997
			DK	620727 T		26-05-1997
			EP	0620727 A		26-10-1994
			ES	2102637 T		01-08-1997
			GR	3024062 T		31-10-1997
			JP	7502735 T		23-03-1995
			US	5387415 A		07-02-1995
			AT	156350 T		15-08-1997
			AT	142484 T		15-09-1996
			AT	181666 T		15-07-1999
			AU	3343093 A		03-08-1993
			AU	679906 B		17-07-1997
			AU	3343293 A		03-08-1993
			CA	2128244 A, C		22-07-1993
			WO	9313753 A		22-07-1993
			WO	9313757 A		22-07-1993
			DE	59303759 D		17-10-1996
			DE	59307073 D		11-09-1997

INTERNATIONAL SEARCH REPORT

Information on patent family members

Internati Application No

PCT/US 00/20484

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
US 5578307 A		DE 59309678 D	05-08-1999
		DK 621775 T	06-10-1997
		DK 621777 T	21-10-1996
		EP 0621775 A	02-11-1994
		EP 0621777 A	02-11-1994
		EP 0701815 A	20-03-1996
		ES 2108258 T	16-12-1997
		ES 2092808 T	01-12-1996
		ES 2133633 T	16-09-1999
		GR 3021223 T	31-01-1997
		GR 3024529 T	31-12-1997
		GR 3030675 T	30-11-1999
		JP 7502736 T	23-03-1995
		US 5876754 A	02-03-1999
		US 6103269 A	15-08-2000
		US 5405616 A	11-04-1995
		US 5401502 A	28-03-1995
WO 9733971 A	18-09-1997	AU 2212397 A	01-10-1997